Tumor Heterogeneity

Tumor Heterogeneity and Permeability as Measured on the CT component of PET/CT Predict Survival in Patients with Non-Small Cell Lung Cancer
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Disclosure of Potential Conflicts of Interest
Balaji Ganeshan and Kenneth Miles have a commercial interest in the tumor textural analysis software through the company TexRAD. There are no other author disclosures

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Tumor Heterogeneity

Translational Relevance

In patients with non-small cell lung cancer (NSCLC) there is need for prognostic factors. This manuscript presents data that shows that tumor heterogeneity as measured on the CT component of PET/CT is a strong independent predictor of survival in patients with NSCLC. Measuring CT tumor heterogeneity could help risk stratify patients with NSCLC for (neo) adjuncts and tumor recurrence monitoring.
Tumor Heterogeneity

ABSTRACT

Purpose: We prospectively examined the role of tumor textural heterogeneity on positron emission tomography/computed tomography (PET/CT) in predicting survival compared to other clinical and imaging parameters in non small cell lung cancer (NSCLC) patients.

Experimental Design: The feasibility study consisted of fifty-six assessed consecutive NSCLC patients (32-males; 24-females; mean-age 67±9.7y) that underwent combined fluorodeoxyglucose (FDG)-PET/CT. The validation study population consisted of sixty-six prospectively recruited consecutive consenting NSCLC patients (37-males; 29-females; mean-age 67·5±7·8y) that successfully underwent combined FDG-PET/CT-dynamic contrast enhanced (DCE)-CT. Images were used to derive tumoral PET/CT textural heterogeneity, DCE-CT permeability, and FDG uptake ($SUV_{max}$). The mean follow-up period was 22.6±13.3 months and 28.5±13.2 months for the feasibility and validation studies. Optimum threshold was determined for clinical stage and each of the above biomarkers (where available) from the feasibility study population. Kaplan-Meier analysis assessed the ability of the biomarkers to predict survival in the validation study. Cox’s-regression determined survival factor independence.

Results: Univariate analysis revealed that tumor CT derived heterogeneity ($p<0.001$), PET derived heterogeneity ($p=0.003$), CT derived permeability ($p=0.002$) and stage ($p<0.001$) were all significant survival predictors. The thresholds used in this study were derived from a previously performed feasibility study. Tumor $SUV_{max}$ did not predict survival. Using multivariable analysis, tumor CT textural heterogeneity ($p=0.021$), stage ($p=0.001$), and permeability ($p<0.001$) were independent survival predictors. These predictors were independent of patient treatment.

Conclusions: Tumor stage and computed tomography-derived textural heterogeneity were the best predictors of survival in NSCLC. The use of computed tomography-derived textural heterogeneity should assist the management of many patients with NSCLC.
Tumor Heterogeneity

INTRODUCTION

Non-small-cell lung cancer (NSCLC) is a common malignancy with a poor prognosis. There is need of predictive factors to refine the management of these patients, e.g., guide the use of surgical adjuncts and help determine patients that are at risk of early reoccurrence, requiring intense monitoring and follow-up (1–4). Current NSCLC guidelines (2–4) reflect survival data based on multiple factors such as performance status (5, 6) and staging (7). Other factors such as tumor metabolism and vascularity have been proposed as prognostic indicators (8, 9). Imaging patients with NSCLC with positron emission tomography (PET) and computed tomography (CT) provides important staging information. In addition these imaging techniques can be used to derive tumor glucose metabolism and vascularity (8, 9). This functional data may in turn be used to predict patient outcome.

Recently researchers have investigated tumor heterogeneity in the search for oncological prognostic markers and mechanisms. Indeed the importance of such an approach has recently been highlighted, by demonstration of genomic tumor heterogeneity with significant implication for treatment and resistance (10). However such an approach is both time consuming and costly. A potentially easier approach is to assess tumor heterogeneity using imaging. These methods assess how grainy or coarse a tumor appears on imaging. PET and CT have both been used to derive tumor textural information and the appearances have been shown to relate to patient outcome in esophageal and colorectal cancer (11-13).
Tumor Heterogeneity

In this study we examine the prognostic potential of tumoral textural analysis using PET/CT in patients with NSCLC compared to tumor staging and other imaging prognostic factors: metabolism and vascularity.
Materials and Methods

Patients

This research study used a feasibility data-set of NSCLC patients to derive optimal thresholds for the markers (imaging and clinical) that predicted survival and then prospectively applied these thresholds within this validation data-set of NSCLC patients. Furthermore the two patient cohorts (feasibility and validation) were independent and from different centers.

Feasibility Data-Set

The feasibility study population consisted of fifty-six consecutive NSCLC patients (32-males; 24-females; mean age 67±9.7y) that underwent staging fluorodeoxyglucose (FDG) PET-CT as part of their routine clinical care between April 2006 and November 2006) at a hospital different from the one used for the validation data-set. [14].

Validation Data-Set

The study population was of similar size to the feasibility population and consisted of sixty-six prospectively recruited consecutive consenting NSCLC patients (37-males; 29-females; mean age 67·5±7·8y) that successfully underwent FDG-PET dynamic contrast enhanced (DCE) CT over a 4-year period from the out-patients clinics of two hospitals. Sixteen tumors were squamous-cell carcinomas, 32 were adenocarcinomas and the remaining 18 mixed/non-specific NSCLC (see Table-1 for staging). All the patients were followed-up in the outpatient setting, at least 3-monthly. Histology was performed by needle biopsy/ bronchoscopic biopsy or surgery. Histological diagnosis was made by an expert thoracic histopathologist. All patients gave informed consent and the Local Ethics Board approved this study.
PET/CT Imaging Protocol

Feasibility Data-Set

All images had been acquired using a PET/CT system with both CT and PET data acquired in one procedure in accordance with a standardized protocol as published [14].

Validation Data-Set

Following a 6h patient fast, images were acquired 1h after injecting 370 MBq of $^{18}$F-FDG using a PET/CT (VCT-XT-Discovery, GE-Healthcare, Waukesha, WI). A CT was performed (for attenuation correction) using $64 \times 3\cdot75$-mm detectors, a pitch of 1·5 and a 5-mm collimation (140 kVp and 80mA in 0·8s). Maintaining the patient position, a whole-body $^{18}$FDG PET emission scan was performed and covered an area identical to that covered by CT (4-min/bed position). Transaxial emission images of 3·27mm thickness (pixel-size 3·9mm) were reconstructed using ordered subsets expectation maximization with two iterations and 28-subsets. The axial field of view was 148·75mm, resulting in 47 slices/bed-position. Next, maintaining the patient position, a DCE-CT was performed during shallow respiration. Patients received 50ml of intravenous contrast medium Iohexol (350 mg/ml iodine, GE Healthcare; Chalfont, UK; 25ml at 5ml/s), at 4ml per second, whilst twelve CT images (1 per 5s with a 5s delay) were acquired axially at 100-150 mAs, 80KV using $64 \times 0\cdot6$-mm detectors (4cm detector coverage).

PET Image Data analysis

In each of the feasibility and validation data-sets, $^{18}$FDG analysis was performed by a nuclear medicine physician with >4 years’ experience of region of interest (ROI) with PET/CT. PET/CT images were displayed conventionally on a proprietary workstation. An automated ROI was
Tumor Heterogeneity

drawn around the tumor and FDG uptake was expressed as the maximum Standardized Uptake Value (SUV$_{\text{max}}$). Automation was performed using a standard ROI analysis tool provided with the scanner, using a threshold method - 42% of the max value (Figure 1) (15, 16).

**DCE Image Analysis**

In the validation data-set, DCE-CT data was analyzed using proprietary CT perfusion software (Winfun, Cambridge Computed-Imaging, Bourne, UK) under the supervision of an operator with 20 years’ DCE-CT experience. On an axial image, a ROI was drawn freehand around the lung tumor within the boundaries of the mass, using PET images for guidance. Vascularity was measured at a single mid-tumor level and a manual correction was made for respiratory motion. A further ROI was drawn freehand around the aorta within the boundaries of the vessel. The tumor flow extraction product - a measure of tumor vascular leakiness (permeability) was calculated using Patlak analysis (17).

**Texture Analysis**

CT Texture Analysis (CTTA)

In each of the feasibility and validation data-sets, CTTA was performed from the attenuation correction images acquired for PET/CT, using a proprietary software algorithm (18) (see appendix for detailed). The operator who performed the CTTA in the feasibility study (had six years’ experience in CTTA) was different from the operator who performed the CTTA in the validation study (chief CT technologist with >10 years of ROI experience under supervision from a researcher with six years’ experience in CTTA). All operators were blinded to all other data. A single 2-dimensional lung CT slice with the largest cross-section area of tumor was used for CTTA. A ROI was drawn accurately contouring the tumor (Figure 2A). This followed similar
Tumor Heterogeneity guidelines as for PET image selection/analysis. The tumor heterogeneity was measured selectively at different texture scales - fine, medium and coarse features (Figure 2B-D) using entropy (a measure of irregularity). Texture values were further normalized (12) to the coarsest scale to give a series of relative texture values (i.e. relative contribution to overall texture made by texture components of different scales) - see appendix.

PET Texture Analysis (PTA)

In the validation dataset, PTA was performed on the SUV images used to measure the SUV$_{\text{max}}$. The images (individual pixel values) with initial units of uptake in Bq/ml were converted (scaled) to SUV calibrated by patient weight and actual tracer activity (taking into consideration the initial tracer activity, amount of decay between the tracer measured time and scan time with respect to the half-life period of$^{18}$F-FDG = 109.8 minutes) with final units of uptake in g/ml. The tumor heterogeneity was measured only on the SUV image without image filtration using entropy (a measure of irregularity – similar in quantification to CTTA). Image filtration was not appropriate owing to the inherently poor resolution of PET (SUV) data.
Statistical Analysis

Statistical analyses were performed using SPSS for Windows version 16.0. The relationships of tumor heterogeneity (CTTA – CT Texture Analysis & PTA – PET Texture Analysis), Stage, $\text{SUV}_{\text{max}}$, permeability, and treatment with patient survival were assessed using Kaplan-Meier (KM) survival analysis. In the case of CTTA, Clinical stage and $\text{SUV}_{\text{max}}$, the optimal thresholds (cut-offs) were determined that best separated the survival plots (poor and good prognostic groups) in the feasibility data-set. These optimal thresholds (cut-offs) were then evaluated in the validation data-set (to test the robustness of the biomarker) using KM survival analysis. In the case of PTA, permeability and treatment, their relationship with patient survival was directly assessed using KM survival analysis in the validation data-set. Differences between KM survival curves for patients above and below each threshold were evaluated by a non-parametric log rank test (p values < 0.05 were considered significant) (18). Multivariate Cox regression was used to determine which parameters were independent predictors of survival (along with the hazard ratio and the confidence interval) and their interactions with treatment analyzed in the validation data-set.
Results

Feasibility data-set

The mean follow-up period was 22.6±13.3 months. The mean survival was 24.1 months, whereas median survival was 28 months. 27 of 54 patients died within 30 months of their PET-CT. The shortest survival time was 1 month. Mean (range) tumor $SUV_{\text{max}}$ for all patients was 14.5 (3.4-37.1) (14).

Validation data-set

The mean follow-up period was 28.5±13.2 months. The mean survival was 25.8 months, whereas median survival was 20 months. 29 of 66 patients died within 30 months of their PET-CT. The shortest survival time was 1 month. Mean (range) tumor $SUV_{\text{max}}$ for all patients was 13.7 (2.1-34.0). A total of 32 of the 66 patients proceeded to radical therapy; 30 underwent surgical resection and two had radical radiotherapy.

Kaplan Meier Analysis - Whole Cohort Analysis

The feasibility data-set (described above) identified the optimal threshold for CTTA, clinical stage and $SUV_{\text{max}}$ at which these markers were the best predictor of survival using univariate analysis using Kaplan Meier (Table 2, Figure 3). In the validation data-set, the strongest predictor of survival was tumor textural heterogeneity ($p<0.001$) measured on CT (CTTA at normalized entropy, medium/coarse texture-scale corresponding to 1.5/2.5, Table 3, Figure 4) and staging ($p<0.001$, Table 3, Figure 4) based on the optimal threshold derived from the feasibility data-set. Tumor maximum standardized uptake value (at the optimal threshold derived from the feasibility data-set) was not a predictor of survival (Table 3). Additionally the
Tumor Heterogeneity

univariate analysis using Kaplan Meier showed that tumor texture heterogeneity measured on PET, radical therapy (Figure A2, appendix) and permeability (Figure 4) were also significant predictors of NSCLC patient survival (Table 3).

Kaplan Meier Analysis - With Radical Treatment as Stratum
In the curative intent group tumoral heterogeneity on CT (CTTA at normalized entropy, medium/coarse texture-scale) was the only factors that were shown to be associated with survival. In the palliative group, tumoral heterogeneity on CT (normalized entropy, medium/coarse texture-scale), stage, and permeability were associated with survival (see Table 3).

Multivariate Analysis - Cox’ Regression
Cox regression analysis indicated that tumoral heterogeneity on CT (CTTA), stage and permeability were independent predictors (CTTA: hazard ratio, HR=412.241, 95% CI 1.45 - 103.05, p=0.021; Clinical stage: HR=5·02, 95% CI 1·89 – 13·32, p=0.001; Permeability: HR=6·01, 95% CI 2·34 – 15·41, p<0·001), while tumor heterogeneity on PET (PTA) and radical therapy were not significant predictors of survival (Table 4). Also there was no significant interaction between the above significant predictors and treatment on overall patient survival.
Tumor Heterogeneity

Discussion

We show in a prospectively collected population of NSCLC patients that clinical stage, vascularity, heterogeneity (on both the CT and PET components of PET/CT) are all significant predictors of patient survival using univariate analysis. Using multivariable analysis tumor heterogeneity (CTTA), permeability and stage were all found to be independent predictors of survival. The threshold level for these markers was determined from a previous feasibility study. Given that performing textural analysis is simple and almost all NSCLC patients undergo CT, these finding have potential management implications for these patients undergoing either radical or palliative therapy.

Prognostic factors are an essential requirement in the management of NSCLC and this is reflected in recent changes in the TNM staging (7), where each sub-group is indicative of outcome. However, despite such refinements, there remains uncertainty and more predictive data is required. This is particularly true in Stage 3A disease where outcome remains variable (4). Moreover, the selection and benefit of surgical patients for neo-adjuvant/ adjuvant treatment is also unclear (20–24). It is important that the possible survival benefits of chemoradiation are balanced by the adverse effects of toxicity to the patient. Moreover, adjunctive regimes are expensive and there may be need for justification by the healthcare provider.

The role of systemic therapy in palliative regimes is also evolving. The rationale for selection of molecular targeted therapy and second line treatment is in need of refining (2, 4, 25, 26). The role of prognostic factors in determining selection is yet to be determined. Finally there is controversy in respect to the need for disease reoccurrence monitoring (4). The use of tumor
heterogeneity may also help identify those patients at particular risk for relapse and thus would benefit from more intensive observation and follow-up.

Why tumoral heterogeneity is related to survival is unclear. It would also be interesting to investigate whether tumor heterogeneity could be related to clonal dominance. It has recently been that there is genomic heterogeneity with in tumors with significant implication for Darwinism theories of tumor resistance (10). Whether textural heterogeneity on imaging relates to underlying genomics would be important to investigate. Given the challenges and expense of measuring tumor genomic signatures, then imaging may be more viable option.

Another possible avenue worthy of further explanation is the possible relationship between tumor heterogeneity and hypoxia. It has been recently shown that tumor textural analysis was associated with tumor hypoxia on histological examinations from NSCLC patients who were administered intravenous pimonidazole prior to surgery (27). Hypoxia is a recognized marker of poor outcome, and as such, a positive relationship between tumor hypoxia and tumor heterogeneity would be biologically consistent.

Imaging derived survival predictors are recognized in NSCLC; the most established being staging data. Tumor uptake of FDG (standardized uptake value or SUV) on PET has been suggested a strong predictor of survival in many studies including a recent meta-analysis (28). PET is part of the diagnostic pathway for NSCLC in those being considered for radical therapy and tumor FDG uptake is relatively easy to measure. As such PET SUV measurements are a potentially clinically viable option. However, it has been recognized that there are difficulties in
assessing whether SUV is an independent survival factor in NSCLC (28). Nonetheless a recent meta-analysis showed tumor SUV was not an independent predictor of outcome (8).

In addition to assessing SUV values, PET textural analysis may be useful and it has been shown to predict survival in esophageal cancer (11). There is a lack of survival data in lung cancer using PET texture, so our finding that survival was not an independent survival factor using PET textural was interesting. However, measuring heterogeneity uptake of $^{18}$FDG on PET is a clinically important parameter to measure as it is can be used to delineate tumor volumes for radiotherapy planning and escalation (boost), such as in colorectal cancer (29). This approach is likely to be useful in NSCLC as well (30).

DCE CT has been shown to predict treatment response in NSCLC (9), but our data is the first to demonstrate a survival association. We also show that it is a significant independent predictor of survival. However, the disadvantage of this approach is the dynamic CT protocol required, is yet to be routinely adopted in clinical practice and there are technical challenges that need to be overcome (31, 32). DCE-CT also entails an additional radiation dose which may need to be as high as 30mSv for reliable whole tumor assessments [32]. In contrast, measuring textural heterogeneity has the advantage of being relatively easy to perform and the textural data can be acquired using conventional CT protocols which are routinely performed as part of standard of care in patients with NSCLC. Furthermore the use of relative texture analysis allows the effect of variations in acquisition parameters (between the feasibility and validation data-sets) on lung tumor texture to be minimized, therefore making this approach applicable across centers.
Study limitations include a limited size population. However, it should be appreciated that all these patients had a substantial array of imaging parameters performed on each individual; SUV\textsubscript{max} from PET, vascularity on DCE CT and textural analysis on conventional CT, and 66 patients is a relatively large population for perfusion CT study, even in the absence of PET. The use of a feasibility population provided a training population to identify the optimal cut-offs for the markers which were further evaluated in the validation population and thereby increased the statistical robustness and we were able to show strong statistical significances. A further possible limitation is the inclusion of patients undergoing both radical and palliative therapy. Yet by having both sets of patients it was possible to show that texture heterogeneity was a survival predictor independent of treatment. Density on CT has been observed as a prognostic factor on CT before e.g. using the presence of air in the tumor (33, 34), however, the use of CT component of PET/CT to measure tumor heterogeneity in lung cancer survival per se is novel.

We chose to perform all image analyses using a single transverse image for each technique. This choice was predicated on the need to match novel image parameters with that most widely proposed as a prognostic marker in NSCLC at the time of study design i.e. SUV\textsubscript{max} which, by quantifying the most FDG-avid pixel, is inherently a single-slice technique. The 4cm cranio-caudal coverage of the DCE-CT was to allow for correction of respiratory motion. DCE-CT assessment of whole tumor would have required even greater coverage with a significantly larger radiation dose. However, single-slice measurements of vascular permeability and heterogeneity could potentially miss areas of greater abnormality at other anatomical levels. Hence, comparison of single- and multi-slice approaches could be included in further validation studies. However, for CTTA, studies to date suggest little difference between these approaches [35].
Finally, there should be awareness of the artifacts caused by motion (breathing) and different slice thickness/resolution between the imaging modalities used in this study.

In conclusion, we show that CT derived tumor heterogeneity and permeability are independent predictors of survival in addition to clinical stage in NSCLC. This finding has potential to aid the management of NSCLC patients with both early and late disease.
Tumor Heterogeneity

References


Tumor Heterogeneity


Tables

Table 1: Lung cancer staging of the patient cohort – a. Feasibility data-set and b. Validation data-set

a.

<table>
<thead>
<tr>
<th>Stage</th>
<th>Number of Patients (Feasibility data-set)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>24</td>
</tr>
<tr>
<td>2</td>
<td>10</td>
</tr>
<tr>
<td>3</td>
<td>14</td>
</tr>
<tr>
<td>4</td>
<td>6</td>
</tr>
</tbody>
</table>

b.

<table>
<thead>
<tr>
<th>Stage</th>
<th>Number of Patients (Validation data-set)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1A</td>
<td>10</td>
</tr>
<tr>
<td>1B</td>
<td>15</td>
</tr>
<tr>
<td>2A</td>
<td>7</td>
</tr>
<tr>
<td>2B</td>
<td>8</td>
</tr>
<tr>
<td>3A</td>
<td>12</td>
</tr>
<tr>
<td>3B</td>
<td>2</td>
</tr>
<tr>
<td>4</td>
<td>12</td>
</tr>
</tbody>
</table>

In the surgical patients (n=30), T and N stage was determined histologically. In the other patients, and for the M stage, the staging was determined by full review of all available data at the multidisciplinary team meeting.
Table 2: Summary of KM survival analysis for the prognostic factors in the feasibility data-set (at the optimal-threshold)

<table>
<thead>
<tr>
<th>Tumor characteristic</th>
<th>Threshold (Cut-Off)</th>
<th>Overall – Mean survival in months (number of patients)</th>
<th>Hazard Ratio</th>
<th>95% Confidence Limits</th>
<th>Overall p – value (KM)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Above Threshold</td>
<td>Below Threshold</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Heterogeneity (CTTA – normalized entropy, medium/coarse scale)</td>
<td>&lt;1.233</td>
<td>34.5 (8)</td>
<td>22.1 (46)</td>
<td>6.110</td>
<td>0.828 – 45.075</td>
</tr>
<tr>
<td>Clinical Stage</td>
<td>&gt;II</td>
<td>17.4 (20)</td>
<td>28.2 (34)</td>
<td>2.830</td>
<td>1.310 – 6.100</td>
</tr>
<tr>
<td>SUV_{max}</td>
<td>&gt;22.8</td>
<td>12.0 (4)</td>
<td>24.7 (50)</td>
<td>3.350</td>
<td>0.997 – 11.251</td>
</tr>
</tbody>
</table>

Table 3: Summary of KM survival analysis (overall and sub-group - radical & palliative treatment) for each prognostic factor in the validation data-set (using the threshold from the feasibility data-set)

<table>
<thead>
<tr>
<th>Tumor characteristic</th>
<th>Threshold (Cut-Off)</th>
<th>Overall – Mean survival in months (number of patients)</th>
<th>Hazard Ratio</th>
<th>95% Confidence Limits</th>
<th>Radical – Mean survival in months (number of patients)</th>
<th>Palliative – Mean survival in months (number of patients)</th>
<th>Overall p – value (KM)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Above Threshold</td>
<td>Below Threshold</td>
<td></td>
<td>Above Threshold</td>
<td>Below Threshold</td>
<td></td>
</tr>
<tr>
<td>Heterogeneity (CTTA – normalized entropy, medium/coarse scale)</td>
<td>&lt;1.233*</td>
<td>39.7 (19)</td>
<td>18.2 (47)</td>
<td>&lt;0.001</td>
<td>41.2 (12)</td>
<td>24.5 (20)</td>
<td>0.027</td>
</tr>
<tr>
<td>Clinical Stage</td>
<td>&gt;II</td>
<td>10.8 (26)</td>
<td>33.1 (40)</td>
<td>&lt; 0.001</td>
<td>11.6 (5)</td>
<td>35.0 (27)</td>
<td>0.070</td>
</tr>
<tr>
<td>SUV_{max}</td>
<td>&gt;22.8</td>
<td>11.5 (9)</td>
<td>25.9 (57)</td>
<td>0.948</td>
<td>11.0 (3)</td>
<td>33.0 (29)</td>
<td>0.727</td>
</tr>
<tr>
<td>Permeability</td>
<td>&lt;14-331</td>
<td>30.0 (43)</td>
<td>14.4 (23)</td>
<td>0.002</td>
<td>34.5 (23)</td>
<td>25.0 (9)</td>
<td>0.179</td>
</tr>
<tr>
<td>Treatment (Radical=0, Palliative=1)</td>
<td>&gt;0</td>
<td>17.1 (32)</td>
<td>32.4 (34)</td>
<td>0.002</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

* - Optimal threshold derived from the feasibility data-set
Table 4: Summary of Multivariate Cox regression analysis model comprising each prognostic factor in the validation data-set

a. Variables included in the Cox equation

<table>
<thead>
<tr>
<th>Tumour Characteristic</th>
<th>Hazard Ratio</th>
<th>95% Confidence Limits</th>
<th>p-value</th>
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</thead>
<tbody>
<tr>
<td>Heterogeneity (CTTA – normalized entropy, medium/coarse scale)</td>
<td>12.241</td>
<td>1.454 – 103.053</td>
<td>0.021</td>
</tr>
<tr>
<td>Clinical Stage</td>
<td>5.020</td>
<td>1.892 – 13.320</td>
<td>0.001</td>
</tr>
<tr>
<td>Permeability</td>
<td>6.009</td>
<td>2.343 – 15.410</td>
<td>&lt;0.001</td>
</tr>
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</table>

b. Variables not included in the Cox equation

<table>
<thead>
<tr>
<th>Tumour Characteristic</th>
<th>Score</th>
<th>p-value</th>
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</thead>
<tbody>
<tr>
<td>Heterogeneity (PTA – entropy without filtration)</td>
<td>0.301</td>
<td>0.583</td>
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<tr>
<td>Treatment</td>
<td>0.686</td>
<td>0.407</td>
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<tr>
<td>Heterogeneity (CTTA)*Treatment</td>
<td>0.825</td>
<td>0.364</td>
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<tr>
<td>Heterogeneity (PTA)*Treatment</td>
<td>0.094</td>
<td>0.759</td>
</tr>
<tr>
<td>Clinical Stage*Treatment</td>
<td>1.039</td>
<td>0.308</td>
</tr>
<tr>
<td>Permeability*Treatment</td>
<td>1.037</td>
<td>0.308</td>
</tr>
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</table>

* - indicates the interaction between the covariates
### Tumor Heterogeneity

Table 5 - To accompany survival curves in FIGURE 3

**A**
Number of patients at risk

<table>
<thead>
<tr>
<th></th>
<th>8</th>
<th>8</th>
<th>7</th>
<th>7</th>
<th>&gt;1.233</th>
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</thead>
<tbody>
<tr>
<td>Number of patients at risk</td>
<td>46</td>
<td>32</td>
<td>24</td>
<td>20</td>
<td>&lt;1.233</td>
</tr>
<tr>
<td>Survival (months)</td>
<td>0</td>
<td>10</td>
<td>20</td>
<td>30</td>
<td></td>
</tr>
</tbody>
</table>

**B**
Number of patients at risk

<table>
<thead>
<tr>
<th></th>
<th>34</th>
<th>28</th>
<th>24</th>
<th>22</th>
<th>&lt;= II</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of patients at risk</td>
<td>20</td>
<td>12</td>
<td>7</td>
<td>5</td>
<td>&gt; II</td>
</tr>
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<td>30</td>
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**C**
Number of patients at risk

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<td>30</td>
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Table 6 - To accompany survival curves in FIGURE 4

**A**
Number of patients at risk

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**B**
Number of patients at risk

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**C**
Number of patients at risk

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<td>30</td>
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Tumor Heterogeneity

FIGURES

Figure 1: A) A fused PET/CT Image to show the region of interest analysis (automated thresholding) in order to determine the measurement of maximal tumour FDG uptake ($\text{SUV}_{\text{max}}$). B) A DCE-CT image to show the region of interest analysis in order to determine the measurement of lung tumour permeability surface for the same patient.

Figure 2: A conventional CT image of a patient (from figure 1) with right upper lobe lung lesion (A) and corresponding images selectively displaying fine (B), medium (C), and coarse (D) texture obtained by using filter values of 1·0 (width, 4 pixels), 1·5 (width, 6 pixels), and 2·5 (width, 12 pixels).

Figure 3: Kaplan Meier curves (along with number of patients at risk for each group over time) showing the proportion of patients surviving for A) heterogeneity – CTTA, B) clinical stage, and C) SUV$_{\text{max}}$ in the feasibility data-set (at the optimal-threshold).

Figure 4: Kaplan Meier curves (along with number of patients at risk for each group over time) showing the proportion of patients surviving for A) heterogeneity – CTTA, B) clinical stage, and C) permeability in the validation data-set (using the threshold from the feasibility data-set).
Figure 3B
Tumor Heterogeneity as measured on the CT component of PET/CT Predicts Survival in Patients with Potentially Curable Non-Small Cell Lung Cancer

Thida Win, KA Miles, Sam M. Janes, et al.

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http://clincancerres.aacrjournals.org/content/suppl/2013/07/01/1078-0432.CCR-12-1307.DC2

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